Reference: *Biol. Bull.* **196:** 94–104. (February, 1999)

Ultraviolet Radiation and Distribution of the Solitary Ascidian *Corella inflata* (Huntsman)

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Abstract. The solitary ascidian *Corella inflata* is a common fouling organism in many areas of Puget Sound and the San Juan Archipelago, Washington, USA. Despite its abundance, it is conspicuously absent from areas that receive direct sunlight. Previous work suggests that ascidians in unshaded habitats can be overgrown and killed by algal overgrowth. In this study, we tested the hypothesis that UV irradiation contributes to *C. inflata* distribution by killing individuals exposed to direct sunlight. To test this, we exposed *C. inflata* embryos, larvae, juveniles, and adults to UV irradiation and measured the responses. We also tested for UV-absorbing compounds in larvae, juveniles, and adults. In the laboratory, UV significantly damaged all life stages; the earliest stages were most vulnerable. A 3-week UV exposure significantly shortened adult life span. Juveniles suffered 100% mortality after only 3 days. Tadpole larvae decreased settlement and metamorphosis after 1 day of UV exposure, and embryos exhibited developmental abnormalities after only 30 minutes of exposure. None of the life-history stages had apparent UV-absorbing compounds. Given the vulnerability of this species to UV, we suggest that its unique life-history traits (*i.e.*, time of spawning, brooding behavior, length of larval life) help it persist in its preferred habitat and avoid dispersal into inappropriate, UV-exposed areas.

Introduction

Corella inflata (Huntsman) is a solitary ascidian common throughout Puget Sound, Washington, and in waters off the west coast of British Columbia. It occurs from the intertidal zone to 45 m (Van Name, 1945) but is most abundant on docks and pilings (Young, 1982). Adults, which may reach 5 cm in length, have a thin, transparent outer tunic. This is in marked contrast to the tough, opaque tunic that protects most other solitary ascidians.

Lambert (1968) studied a population of *C. inflata* in a Puget Sound marina for 12 months and observed mass mortality in the early spring. The mortality coincided with a period of heavy diatom growth, and Lambert suggested that smothering diatom mats were responsible for the ascidian deaths. This conclusion was supported by observations that mass mortality occurred only in areas exposed to full solar radiation; *C. inflata* in shaded habitats survived.

Recent research suggests that another factor may contribute to mortality of *C. inflata* in exposed areas. Increasing interest in the status of the stratospheric ozone layer has led to intense study of the deleterious effects of ultraviolet radiation (UV). Jokiel (1980) first demonstrated the damaging effects of UV radiation on tropical marine invertebrates (including ascidians). More recent work has shown that the UV-B portion of the spectrum (280–320 nm) is particularly lethal to marine bacteria, plankton, invertebrates, and fish (reviewed by Worrest, 1982; Hardy and Gucinski, 1989; see also Shick *et al.*, 1991; Karentz *et al.*, 1991; Karentz, 1994a, b). There are indications that marine invertebrate embryos and larvae may be particularly sensitive to solar radiation (Damkaer *et al.*, 1980; Pennington and Emlet, 1986; Biermann *et al.*, 1992; Adams and Shick, 1996).

The habitat (primarily shallow water) and anatomy (thin, transparent tunic) of *C. inflata* may make it particularly vulnerable to UV damage, and the effects may not be limited to adult animals. Unlike most solitary ascidians, *C. inflata* holds its embryos in a spacious brood chamber. The eggs are buoyant and float to the top of the chamber where there is potential for UV-induced damage during development.

Received 26 November 1997; accepted 6 November 1998.

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The purpose of this study was to determine how UV radiation affects *Corella inflata*. We examined field distributions of ascidians to determine whether population density was correlated with UV exposure. We measured the vulnerability of embryos, larvae, juveniles, and adults to UV damage. Finally, we examined larvae, juveniles, and adults for UV-absorbing compounds.

Materials and Methods

Field sampling

We selected docks in two marinas (Anacortes Marina and Skyline Marina) in Anacortes, Washington, for the field portion of this study. These marinas have similar construction designs, with docks that project out into a sheltered embayment. A roof shades inner dock slips, but outer slips are not covered and receive full sunlight. Because the docks rise and fall with the tides, they harbor diverse invertebrate communities including sponges, hydrozoans, polychaetes, barnacles, bivalves, bryozoans, and ascidians (particularly *Corella inflata*). Macroalgae are absent from most docks at both locations.

At each marina, we chose four nonadjacent slips (two shaded and two exposed) for detailed study. Five permanent sampling locations were marked at 2.5-m intervals along each slip. We measured *C. inflata* densities at each location by placing a 25 × 25 cm quadrat on the side of the slip just below the water's surface and counting all *C. inflata* individuals within the quadrat. We made counts in June and again in August, 1994.

To determine how the density of *C. inflata* related to UV exposure, we measured spectral irradiance 5 cm below the water's surface at each quadrat location. Measurements (from 300–700 nm at 2-nm intervals) were made with a LI-COR 1800UW spectroradiometer (see Kirk *et al.*, 1994, for a discussion of the measurement characteristics of this instrument) on a clear sunny day (15 July 1994) at 1300 h. We also measured light attenuation in the marinas by taking spectral scans at 50 cm and 1 m at several exposed quadrat locations. Finally, we went just outside the entrance to the Skyline Marina and measured light at 1-m intervals from the surface to 20 m. This allowed us to determine penetration of UV-A and UV-B in the local waters.

Because we could not measure the complete UV-B spectrum (from 280–320 nm) due to limitations of the instrument, we used a 2nd order regression of the data from 300–320 nm to extrapolate the curves from 280 to 300 nm. We then integrated the data files to get separate measurements of total UV-B (280–320 nm), UV-A (320–400 nm), and PAR (photosynthetically active radiation, 400–700 nm).

Because algal overgrowth could be a confounding factor in our sampling, we monitored algal growth at the study sites. We hung transparent acrylic strips (2-cm wide × 15-cm long) at all quadrat locations. After 6 weeks (1 July–15 August, 1994), they were collected and two 4-cm^2 areas, one at 5 cm and the other at 15 cm, were wiped with a GFF filter. We extracted the filters in 90% acetone for 24 h and made fluorometric readings of chlorophyll *a* (Parsons *et al.*, 1984). The two values were averaged to give a measurement of algal growth at each quadrat location.

Laboratory experiments

To measure the sensitivity of *C. inflata* to UV, we used an enclosed, flow-through seawater tank equipped with two UV bulbs (Q-Panel Company, UVB-313) and two cool-white fluorescent bulbs. A cellulose acetate bulb sheath filtered out wavelengths less than 290 nm. The tank was divided into two sections with nylon netting. The control section was covered by a UV-filtering shield (Atohaas North America, Plexiglas UF3, 3-mm thickness) that reflected wavelengths below 400 nm. To compare light in the two treatments, we measured the irradiance spectra in the shielded and unshielded portions of the tank (at 5-cm depth) with the spectroradiometer.

Adult sensitivity. To determine if UV exposure affects adult *C. inflata*, we placed 20 individuals in 240-ml polypropylene cups from which the bottoms had been removed and replaced with Nitex screen. A slit in a piece of open-cell foam attached to the inside wall of the cups held the ascidians in a normal position (horizontal with the excurrent siphon and brood chamber up; Young, 1988) but was flexible enough to allow normal feeding. The cups were randomly assigned to either UV-exposed ($n = 10$) or shielded ($n = 10$) treatments. Foam collars kept the cups floating with the adults about 3 cm below the surface of the water. The tank was placed on a 15:9 h light/dark cycle and survival was monitored for 22 days. Life spans (up to 22 days) in the two treatments were compared by one-way ANOVA. We set α at 0.05 for all statistical analyses and used Hartley's F_{max} test (Sokal and Rohlf, 1995) to test for homogenous variances before each analysis.

Juvenile sensitivity. We collected larvae by exposing adult *C. inflata* to bright light shortly after collection. Thirty larvae were placed in each of 12 roughened polystyrene petri dishes (100-mm diameter, 15-mm deep) and allowed to settle. Positions of the juveniles were marked with a permanent ink marker on the back of the dishes. We filled the dishes with fresh seawater and randomly assigned them to the UV-exposed ($n = 6$) or shielded ($n = 6$) treatments. Mortality was determined after 4 days of treatment. Because of unequal variances, we used a Kruskal-Wallis nonparametric ANOVA to compare treatments.

Larval sensitivity. Tadpole larvae were exposed to UV radiation to measure effects on settlement. Six petri dishes containing 30 newly released, swimming tadpole larvae were randomly assigned to the UV-exposed ($n = 3$) or

shielded ($n = 3$) sections of the tank. Percent settlement in each treatment was determined after one light cycle (15 h UV exposure). Treatments were compared by one-way ANOVA.

We also subjected tadpole larvae to different periods of UV exposure to determine a damage threshold. Twenty-one replicate petri dishes (with 30 newly released tadpole larvae per dish) were prepared. Three dishes (the 0-exposure control) were placed in the shielded portion of the tank. The other 18 dishes were exposed to the UV light. At 30-min intervals, we moved three randomly chosen plates into the shielded portion of the tank until all plates had been moved over. Twenty-four hours later, percent settlement was determined for all plates. We analyzed the relationship between UV exposure time and percent settlement by simple linear regression.

Sensitivity of developing embryos. Approximately 75 adults were light shocked to obtain fertilized eggs. Eggs from all the adults were mixed and 30 were arbitrarily assigned to each of 21 petri dishes. The developing embryos were exposed to UV radiation in the experimental tank (in groups of three replicate dishes). Exposure intervals were staggered as described above to determine the threshold at which development becomes abnormal. After 24 h, all dishes were examined and the proportion of eggs that had (1) reached the tadpole stage, (2) arrested development at the morula stage, or (3) become abnormal at, or before, the 16-cell stage was determined. We used a chi-square test for independence to determine whether length of UV exposure affected the developmental stage the embryos reached. To avoid sacrificial pseudoreplication (Hurlburt, 1984), we analyzed only one randomly chosen replicate from each exposure period.

Outplants

To determine if recruiting *C. inflata* could survive in exposed areas of the marina where they were not usually found, newly settled ascidians were transplanted into the field. In the laboratory, 30 tadpole larvae were placed in 100-mm diameter petri dishes ($n = 20$) and allowed to settle. Settlement locations were marked on the back side of each dish with a permanent marker and five marked plates were hung vertically, front side out, on four slips in Skyline Marina (two shaded and two unshaded). After 6 days, we compared survival of the juveniles in the shaded and exposed treatments with a Kruskal-Wallis nonparametric ANOVA.

Recruitment

We monitored *C. inflata* recruitment to determine whether larvae ever colonize exposed docks. Six 100-mm diameter cement plates were hung from two shaded and two unshaded slips in Anacortes Marina and left in place from 8 July to 15 August, 1994. We then collected the plates, counted the recruits, and used correlation analysis to test for relationships between irradiance, algal growth, and adult density and number of recruits.

If algal overgrowth causes mortality of *C. inflata* at our site, larvae should avoid settling on algal-covered surfaces (such as those in exposed sites). We tested this by placing 15 roughened petri dishes (100-mm diameter) in Skyline Marina. Three of the dishes hung from a covered dock where darkness prevented algal growth. The remaining 12 dishes hung from a dock exposed to full sunlight. After 2 weeks, the exposed dishes had developed a layer of filamentous algae. We collected all the dishes and divided them among three treatments: no algal cover (3 dishes from the covered dock), 100% algal cover (3 dishes from the exposed dock), and 50% algal cover (9 exposed dishes that we carefully scraped to remove the algae from half of the bottom surface). The dishes were filled with fresh seawater and 30 *C. inflata* tadpole larvae were added to each. After 24 h, settlement in each dish was recorded. Settlement in 100% algal-covered dishes ($n = 3$) was compared to dishes with no algae ($n = 3$) with one-way ANOVA. We used a paired Student's t test to compare larval settlement in clean or algal-fouled halves of the scraped dishes ($n = 9$).

UV-absorbing compounds

To determine whether *C. inflata* has UV-absorbing substances, we extracted compounds from three large adults (between 1.5 and 2.5 cm long), five juveniles (less than 1.0 cm long), and the pooled tadpole larvae from 50–75 adults. The tunics were removed from the adults and analyzed separately from the bodies (we hypothesized that UV-absorbing compounds, if present, would be concentrated in the tunic). The bodies were carefully cleaned to remove all feces and gut material and ensure that only *C. inflata* compounds were measured. Tadpole larvae were collected by light shocking adults. We lyopholized and extracted the samples in 80% methanol as described by Karentz *et al.* (1991). Absorbance of the extracts was measured spectrophotometrically at 2-nm intervals from 280 to 400 nm.

The dry weights of the *C. inflata* samples were unequal (large tunics = 120 mg, large bodies = 100 mg, small tunics = 170 mg, small bodies = 100 mg, tadpole larvae = 120 mg). To permit direct comparison among the samples, we calculated relative absorptivity by the following equation.

$$\text{relative absorptivity} = \frac{\text{Measured absorbance}}{\text{path length (cm)} \times \left(\dfrac{\text{tissue dry weight (mg)}}{\text{solvent volume (ml)}}\right)}$$

This equation is derived from Beer's law (Skoog and

West, 1974) except that absorptivity in Beer's law is the molar absorptivity of a single absorbing substance. Since we do not know the composition of our extracts, we consider absorptivity in this case to be a relative measure.

To test the effectiveness of the tunic as a physical barrier to UV damage, we dissected the tunics from one juvenile and one adult *C. inflata,* placed them directly in the spectrophotometer light path and measured absorbance from 280 to 400 nm. By substituting tunic thickness (≈900 μm) for path length and tunic density ($g \cdot cm^{-3}$) for the concentration measure in the above equation, we calculated relative absorptivities that were directly comparable to those measured with the extracts.

Results

Field sampling

Corella inflata was entirely restricted to shaded slips in both marinas, and densities decreased toward the slip ends where shading was less complete. At both marinas, there was a strong negative relationship between algal growth (as measured by Chl *a* concentration on acrylic strips) and *C. inflata* density and between UV irradiance and *C. inflata* density (Fig. 1). The relationship between *C. inflata* density and UV-B irradiance alone followed the same pattern and had similarly high correlations ($r = -0.88$, $P < 0.001$ for Anacortes Marina and $r = -0.68$, $P < 0.001$ for Skyline Marina). There was an apparent threshold in both irradiance and chlorophyll concentration above which few *C. inflata* individuals occurred. This threshold corresponded to the point at which slips were no longer shaded. There was essentially no change in densities of ascidians between the two sampling dates (June and August).

A vertical profile showed a logarithmic decrease in UV irradiance with depth (Fig. 2). UV-B was detected to 5 m. Levels of UV-B corresponding to threshold intensities for adult distributions in the marinas (Fig. 1) occurred at depths between 1.5 and 2 m. UV-A was measurable to 14 m.

Laboratory experiments

Light levels in our experimental tank differed significantly from those measured in the field. Although total UV-B irradiances were remarkably similar, total UV-A was 30 times lower in our tank than in the field; total PAR was 32 times lower (Table I). Closer examination of individual spectra reveals that tank and field light quality differed within these wavelength ranges. UV in the experimental tank was skewed to the more biologically damaging, low wavelengths (Fig. 3). Without a weighting function for this species, we cannot determine how this affected the experimental animals. It is likely, however, that the tank overemphasized low-wavelength UV-B effects and under-emphasized UV-A and PAR effects. The Plexiglas shield effectively removed all UV-B and UV-A from the shielded portion of the tank. The shield also caused an 8.5% drop in total PAR, with the greatest effect at wavelengths between 400 and 415 nm (Table I, Fig. 4).

Adult sensitivity. At irradiances present in the tank, UV damaged all *C. inflata* life-history stages. After 8 days, exposed adults became opaque and many were dead after 10 days; after 21 days, all exposed animals were dead. Control animals also experienced some mortality, probably due to handling. However, mortality rates were lower and leveled off after 17 days. The average life span of the UV-exposed adults (mean = 13.9 ± 1.2 d SD) was significantly lower than that of the shielded controls (18.0 ± 1.1 d, $F = 5.8$, $P = 0.02$) within the 21 days of the experiment. This result underestimates the true effect because the experiment was terminated at 22 days and many of the control animals were still living.

Juvenile sensitivity. UV exposure affected juveniles even more strongly than it did adults. In the laboratory, juveniles in the exposed treatment showed 100% mortality after only 4 days (controls had 57.0% ± 11% mortality, $H = 9.46$, $P = 0.002$). Similar results were seen in the field outplants. All juveniles had disappeared from dishes in the unshaded habitat after only 3–6 days, whereas 36.6% ± 8.3% of the shaded juveniles were still alive at the end of the experiment ($H = 7.27$, $P = 0.007$). Examination of the dishes in which the juveniles were outplanted revealed minimal algal growth, suggesting that overgrowth was not responsible for the mortality.

Larval sensitivity. Very few *C. inflata* larvae exposed to UV light settled successfully in the laboratory (2.3% ± 0.4% compared to 56.8% ± 0.8% for the shielded individuals, $F = 56.4$, $P = 0.001$). This underestimates the true effect because many larvae that had not settled in the shielded treatment were still actively swimming; all exposed larvae were dead. The effects of exposure were cumulative; as exposure time increased, settlement success decreased (Fig. 5).

Sensitivity of developing embryos. UV interfered with the normal development of *C. inflata* embryos (Fig. 6, χ^2 for one set of replicates = 303.48, $P < 0.001$). Even limited exposure had significant developmental consequences. After only 30 min of exposure, less than 20% of the embryos developed to tadpole stage. As exposure time increased, early cleavages became more abnormal, with many embryos failing to pass the 4- or 8-cell stages. No normal development occurred after 1.5 h of UV exposure.

Settlement and recruitment

Of 199 *C. inflata* individuals recruiting to plates in Anacortes Marina, 181 (90.9%) were on plates on shaded slips. The remaining 18 recruits on exposed plates were

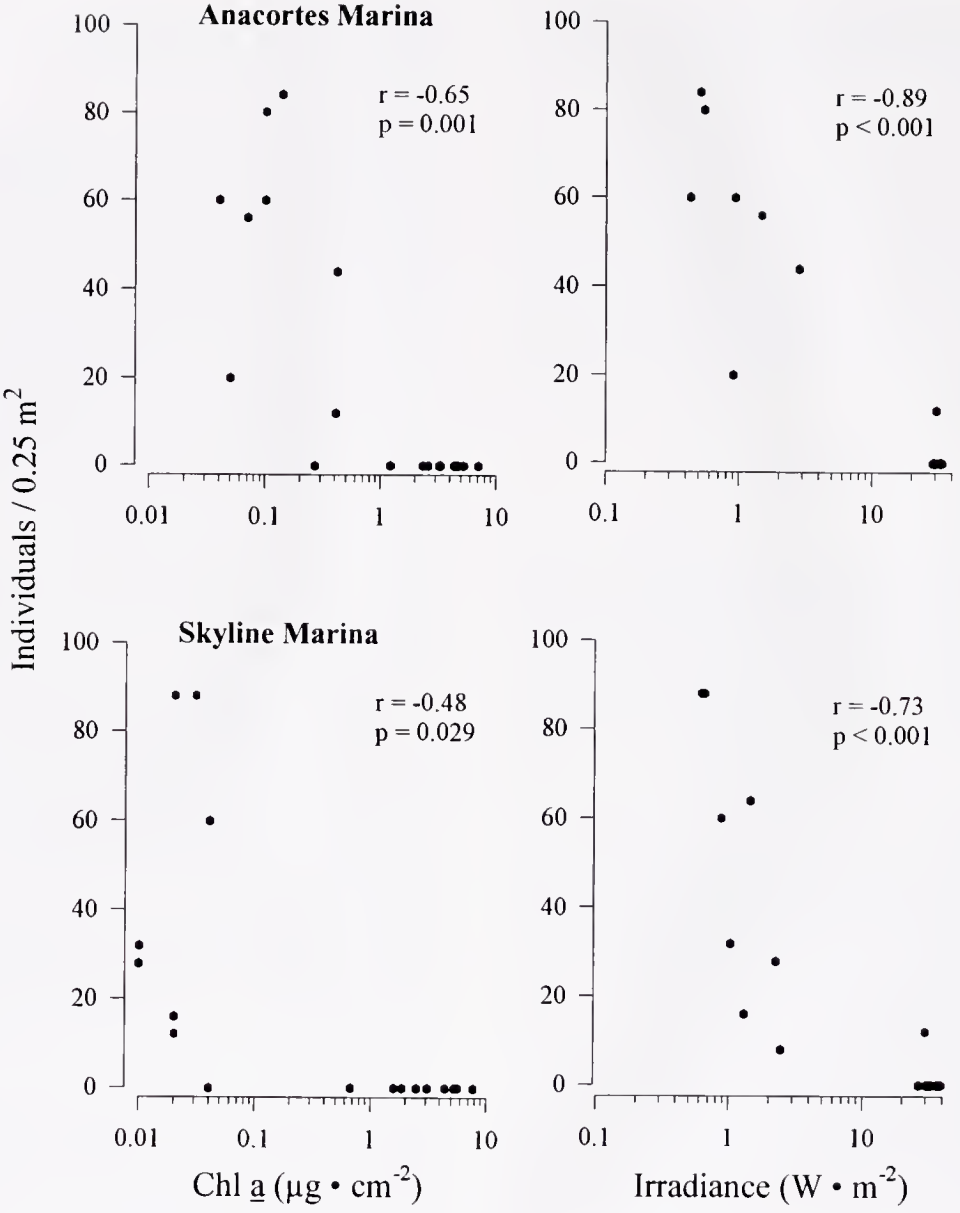

Figure 1. Density of *Corella inflata* in Anacortes and Skyline Marinas. Densities are plotted as a function of Chl *a* concentration (based on overgrowth of clear acrylic strips) and UV irradiance (300–400 nm). Correlation coefficients (*r*) and *P* values are shown.

on the lower edges where they were shaded by the plate itself.

In laboratory assays, a developed algal mat did not affect *C. inflata* settlement. Similar numbers of larvae settled successfully and metamorphosed on clean and algal-fouled surfaces, and larvae showed no preference for clean rather than algal-fouled surfaces when given a choice (Fig. 7). However, because of low replication, the power of these tests was low (β = 0.65 for the single choice and 0.24 for the preference experiment; Cohen, 1988). Levels of algal fouling in the dishes reached 0.08 μg Chl $a \cdot cm^{-2}$.

UV-absorbing compounds

All methanol extracts showed minor absorptivity peaks in the UV range (Fig. 8). Tunic extracts absorbed much less UV than extracts of the bodies. Absorbance was greatest for the bodies of small specimens of *C. inflata*. Tadpole extracts showed a small peak at 290 nm. The peaks (λ_{max}) differed slightly among the samples, but all were between 294 and 300 nm. None fell within the wavelength range in which sunscreening mycosporine-like amino acids normally occur. The tunic provided little physical barrier to UV: absorptivi-

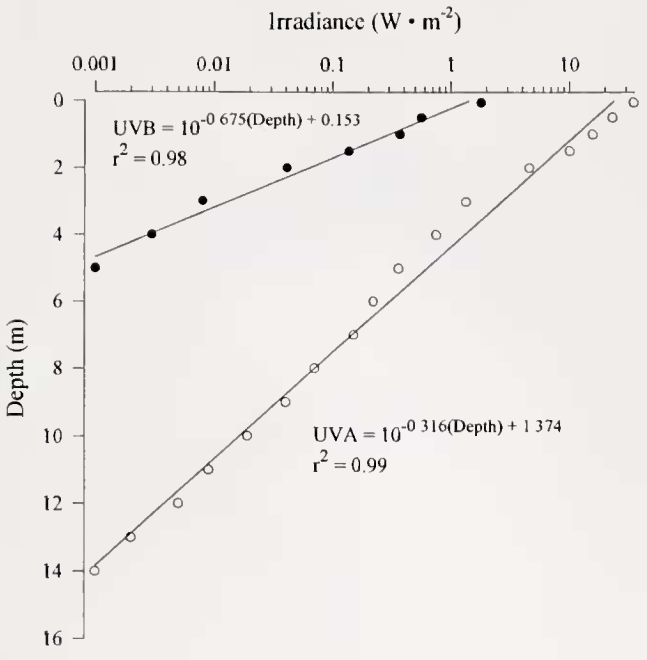

Figure 2. Penetration of UV-B (300–320 nm) and UV-A (320–400 nm) in the water column at the entrance to Skyline Marina. Simple regression lines and equations are shown. Measurements were made at 1300 h on 15 July 1994.

ties were only slightly above those seen in the chemical extracts (Fig. 8). For comparison, we calculated the relative absorptivity of mylar (a synthetic material that filters wavelengths <315 nm). Mylar relative absorptivity at 294 nm was 0.34.

Discussion

Unlike many Puget Sound ascidians, which occur primarily in subtidal benthic habitats, *Corella inflata* is found in greatest abundance on floating docks (Young, 1982). It is not normally found on fixed intertidal structures because it does not tolerate desiccation. The dock habitat may be a refuge from benthic predators that prefer this species to other ascidians that have thicker, heavier tunics (Young, 1985). However, although these artificial substrates may provide protection from predators, they expose *C. inflata* to hazards associated with high levels of solar radiation.

Young and Chia (1984) outplanted juveniles of *C. inflata* to subtidal locations at 4.5-m depths in shaded and unshaded dishes. Mortality was significantly higher in the unshaded dishes, presumably due to algal overgrowth (though this is still within the range to which UV penetrates; Fig. 2). Since algal overgrowth apparently can kill juveniles, we predicted that larvae should detect and avoid algal-filmed surfaces, particularly since larvae of many ascidian species show strong settlement specificity (reviewed by Svane and Young, 1989). In laboratory testing, however, larvae settled readily on surfaces that were covered with filamentous algae, sometimes attaching directly to the algae themselves.

Goodbody (1963) and Goodbody and Gibson (1974) outplanted juvenile *Ascidia nigra* (another solitary ascidian) to the field at depths of 1.2 and 2.1 m and measured cohort survival. They found very high mortality, particularly among individuals at the shallower depth. Mortality was significantly lower in shaded treatments, particularly at the 1.2-m depth. The authors hypothesized that one source of the mortality was smothering from accumulation of benthic diatoms or filamentous algae in the unshaded treatments. The mortality of the outplanted cohorts was highest within the first 15 days. Interestingly, the deep black pigmentation characteristic of adult *Ascidia nigra* does not develop until about the 15th day after larval settlement, and the authors noted that mortality decreased sharply after the pigment appeared (Goodbody and Gibson, 1974). Our results with *C. inflata* suggest that UV could be an alternative source of mortality for young ascidians in exposed habitats.

If the habitat of *C. inflata* makes this species vulnerable to UV damage—as suggested by our results—we expect it to show adaptations to avoid UV exposure in shallow-water habitats. Many organisms possess UV-absorbing, mycosporine-like amino acids (MAAs) that may prevent UV damage (reviewed by Karentz, 1994a). The Antarctic ascidian (*Molgula enodis*) contains seven MAAs (Karentz *et al.*, 1991) with distinct absorbance peaks around 330 nm; the Western Pacific *Halocynthia roretzi* contains a UV-absorbing sunscreen that absorbs maximally at 337 nm (Kobayashi *et al.*, 1981).

C. inflata absorptivities peaked between 290 and 300 nm, suggesting that MAAs are not responsible for the UV absorbance we measured. The body of this ascidian contains uric acid crystals that are deposited there as metabolic waste products (Lambert *et al.*, 1998). It is possible that uric acid provides some limited protection from UV damage (a uric acid absorbance peak occurs at 292 nm). Another possibility

Table I

Comparison of UV-B (300–320 nm), UV-A (320–400 nm), and PAR (400–700 nm) in Skyline Marina, Anacortes, WA, where Corella inflata *populations were monitored (15 July 1994) and in the tank where experiments were done (compare Fig. 3)*

Location	Depth (cm)	Intensity (W • m^{-2})* UV-B	UV-A	PAR
Tank				
UV-exposed	5	1.10	1.12	9.99
Shielded	5	0.05	0.06	9.14
Marina	5	1.80	34.59	328.70
	50	0.56	23.02	248.00
	100	0.37	15.68	226.60

* Measured with a LI-COR 1800UW spectroradiometer.

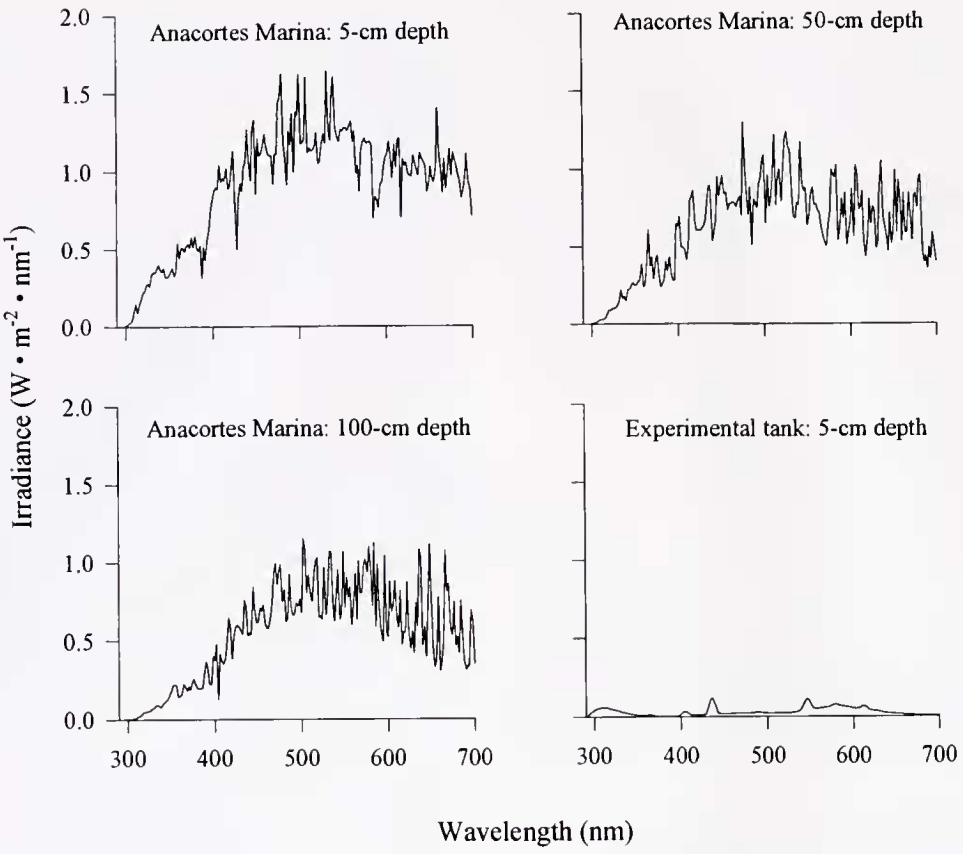

Figure 3. Irradiances at 5, 50, and 100 cm in Skyline Marina and in the flow-through seawater tank used for laboratory experiments (5-cm depth). Due to instrument constraints, we were unable to measure the UV-B below 300 nm. Lines on the figures between 290 and 300 nm are extrapolated second-order regressions from the data for 300 to 320 nm.

is that the absorbance we saw came from cell secretions known as "ornaments" (Cloney, 1990). Embryonic test cells produce ornaments that are deposited on the larval tunic of *C. inflata* and other ascidians (Cloney and Cavey, 1982; Cloney, 1994). The ornaments are composed largely of opal, a form of silicon dioxide (Monniot *et al.,* 1992). It is unlikely that the opal alone absorbs UV, but if the ornaments contain other UV-absorbing substances, they might provide larvae with some protection (R. A. Cloney, pers. comm.).

With little structural or chemical protection, *C. inflata* appears vulnerable to UV damage in all life-history stages. Embryos were the most vulnerable. Even short exposures caused developmental abnormalities; after only 30 min of laboratory UV exposure, many embryos arrested at the morula stage. This agrees with Jeffrey (1990), who also found that UV-irradiated ascidian embryos failed to gastrulate. Although embryos were most sensitive to UV, larvae, juveniles, and adults were also affected. We suggest that the unique life-history characteristics of *C. inflata* allow the populations to persist in shallow habitats despite this vulnerability.

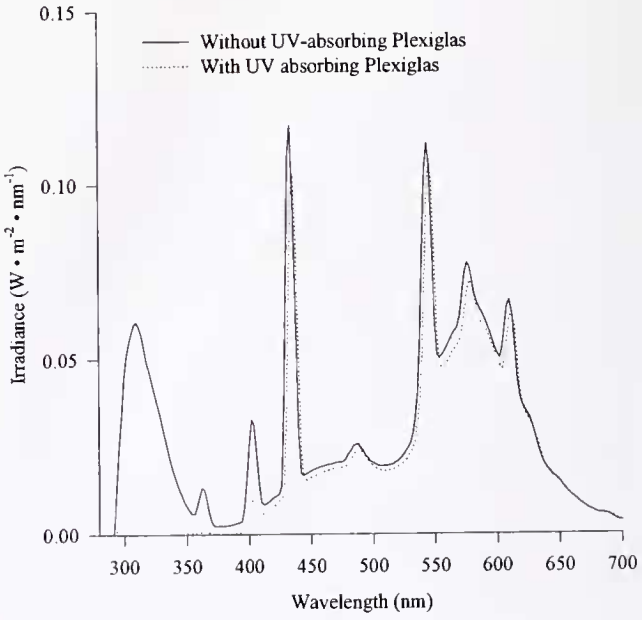

Figure 4. Irradiance spectra in the shielded and unshielded treatment portions of the experimental tank. Wavelengths from 290–300 nm are extrapolated second-order regressions from the data for 300 to 320 nm.

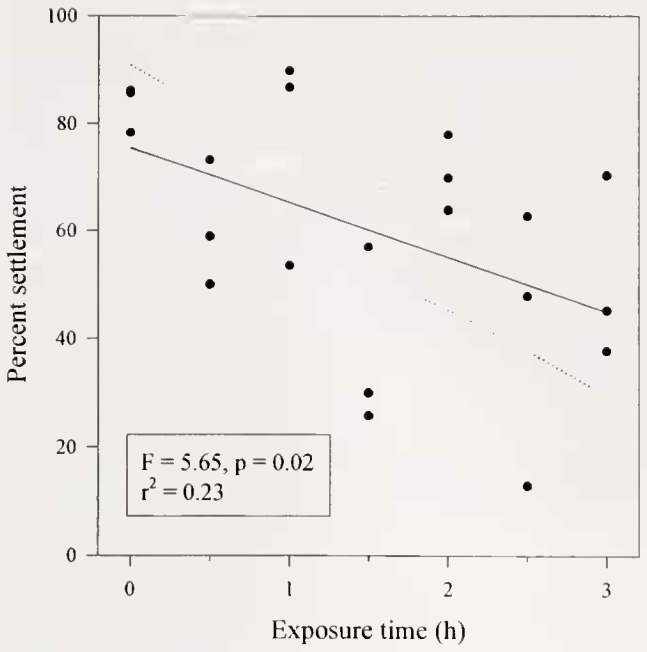

Figure 5. Cumulative effects of UV exposure on *Corella inflata* settlement. Exposures ranging from 0 to 3 h in 30-min intervals. A simple linear regression with a 95% confidence interval and regression statistics are shown.

C. inflata is one of a small number of solitary ascidians that brood their developing embryos. The hermaphroditic, self-fertile adult releases eggs just after dawn (Lambert *et al.*, 1995). If the eggs were freely released into the water, their ammonium-filled follicle cells (Lambert and Lambert, 1978) would carry them directly to the surface, where they would be exposed to full daytime sunlight during their development. Assuming that UV effects in the field are similar to those measured in the laboratory, our study suggests that this exposure could be fatal to the developing embryos.

Instead, *C. inflata* possesses an inflated atrium that functions as a brood chamber (Child, 1927). The horizontal, atrium-up position of the adults on the docks favors retention of the embryos within the chamber (Lambert, 1968; Young, 1988). The floating embryos hatch within the brood chamber after 24–26 h (Lambert *et al.*, 1995). Thus, the most vulnerable life stage is protected within the adult. Because adults persist only in shaded environments, this is a particularly safe environment for the embryos, even given the limited UV protection the adult tunic provides. Interestingly, *Corella willmeriana* and *Corella parallelogramma*, both free spawners, also have floating eggs (Hüus, 1939; Lambert *et al.*, 1981). Perhaps their early embryos are more resistant to UV damage than are those of *C. inflata*.

Until recently, it was thought that tadpoles were released from adult *C. inflata* soon after hatching. However, given that the eggs are produced in early morning and take about 24 h to develop, the larvae would be entering the environment when UV damage would be greatest, particularly since the larvae are apparently geonegative and swim toward the surface (Young, 1988).

Olson (1983) noted that larvae of the ascidian *Didemnum molle* are released near midday, when light levels are highest. This species contains a symbiotic cyanophyte (*Prochloron*) that is sensitive to both high and low light levels. Midday release permits larvae to choose an appropriate habitat while light levels are at their maximum. Newly released larvae swim briefly toward the surface, then descend to the bottom and settle (dispersal times were less than 10 min; Olson, 1983). The exposure time may be sufficiently brief, or the larvae sufficiently protected, that damage from UV does not occur.

C. inflata larvae can detect light but do not show phototaxis (Young, 1988), nor does light have a strong effect on settlement behavior (Young and Chia, 1985). Furthermore, larvae are incapable of detecting wavelengths below 425 nm (Young, 1982). Given these limitations, the larvae may be unable to use light as a cue to an appropriate settlement site (*i.e.*, one in which the sessile juvenile will not be killed by UV exposure).

Lambert *et al.* (1995) suggest that *C. inflata* tadpoles,

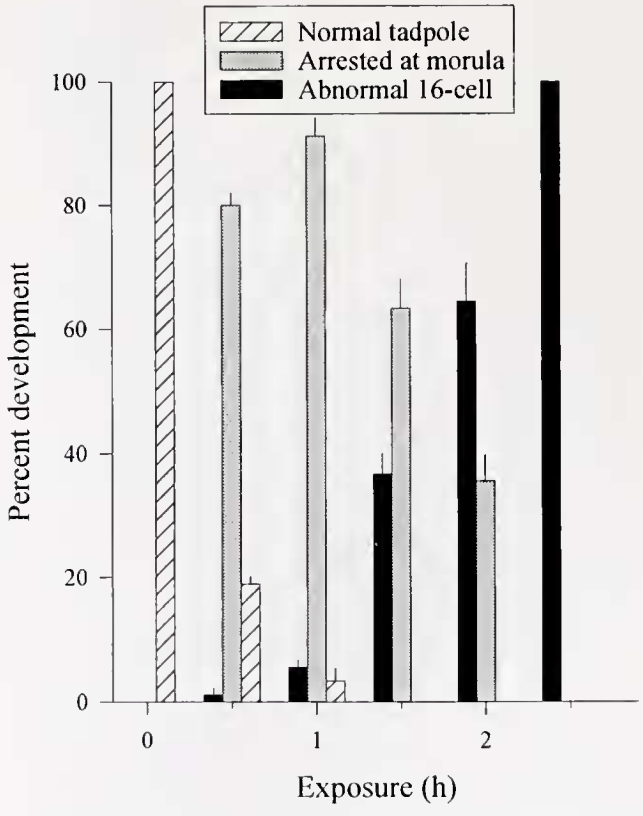

Figure 6. Effects of UV exposure on *Corella inflata* embryonic development. Morula stage is a solid ball of cleaved cells (probably 32–64 cells in this case). Standard errors are shown.

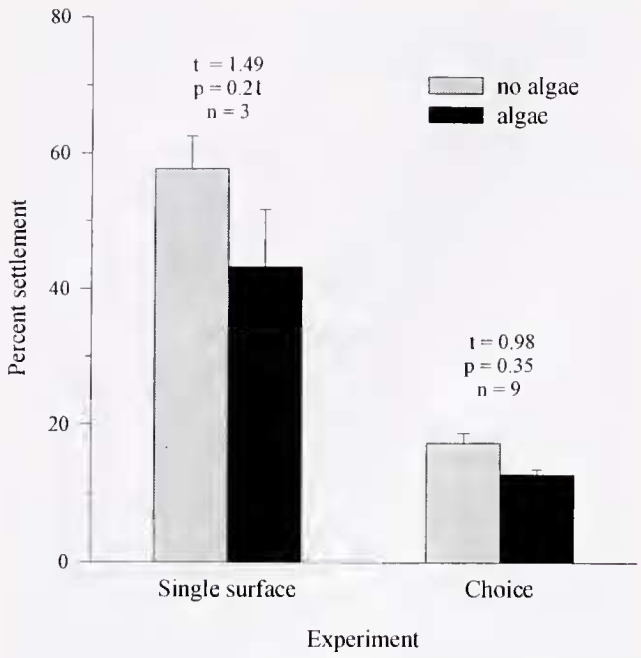

Figure 7. Effects of filamentous algae on *Corella inflata* settlement. There were no differences in settlement success on clean and fouled surfaces, nor did larvae choose clean surfaces over algal-fouled when given a choice (standard errors are shown).

which hatch in the early morning, are retained within the adult atrium for 12–17 h and released at night (they found swimming tadpoles in the atria of individuals collected at 2130 h). The larvae are competent to settle when released and probably settle near the adult that released them. Lambert (1968) described a settlement pattern of juvenile *C. inflata* clumped around adults.

Local settlement after a very brief planktonic period (in the dark) may improve the chances of survival for this species, which is apparently vulnerable to, but cannot detect, UV irradiation or algal overgrowth. This scenario is supported by our observation that, though adult *C. inflata* were abundant on the shaded undersides of exposed docks, no recruitment occurred to plates attached to the unshaded dock sides. Such highly localized recruitment has been described for a number of ascidian species (reviewed by Svane and Young, 1989). Note, however, that Cohen (1990) found significant genetic variability in *C. inflata* despite its self-fertility and short larval life. Outcrossing suggests that some dispersal of eggs or larvae is taking place.

C. inflata generally appears to follow a developmental scheme in which (1) embryos develop within the atrium of an adult in a shaded environment, (2) larvae hatch within the brood chamber and remain there throughout the daytime when UV effects are strongest, (3) competent larvae are released at night, and (4) the larvae quickly settle near the parent. This life-history pattern may allow *C. inflata* to opportunistically exploit floating-dock habitats in which other ascidians could not survive. For example, the range of *Corella willmeriana* overlaps that of *C. inflata*. These species are morphologically similar and have been confused by a number of authors (see Lambert *et al.*, 1981, for a review). However, *C. inflata* is found primarily on docks and pilings, whereas *C. willmeriana* is almost entirely restricted to deeper subtidal habitats; it is rare on docks (Lambert *et al.*, 1981). This distributional dichotomy is accompanied by a significant morphological difference: *C. willmeriana* does not have an inflated atrium and does not brood its young. The absence of these features may prevent it from invading the habitat *C. inflata* has successfully colonized.

At present, we do not know the relative importance of UV-B and UV-A to *C. inflata* distributions. The more damaging UV-B wavelengths penetrate to depths at which *C. inflata* occurs. However, UV-A intensities are much

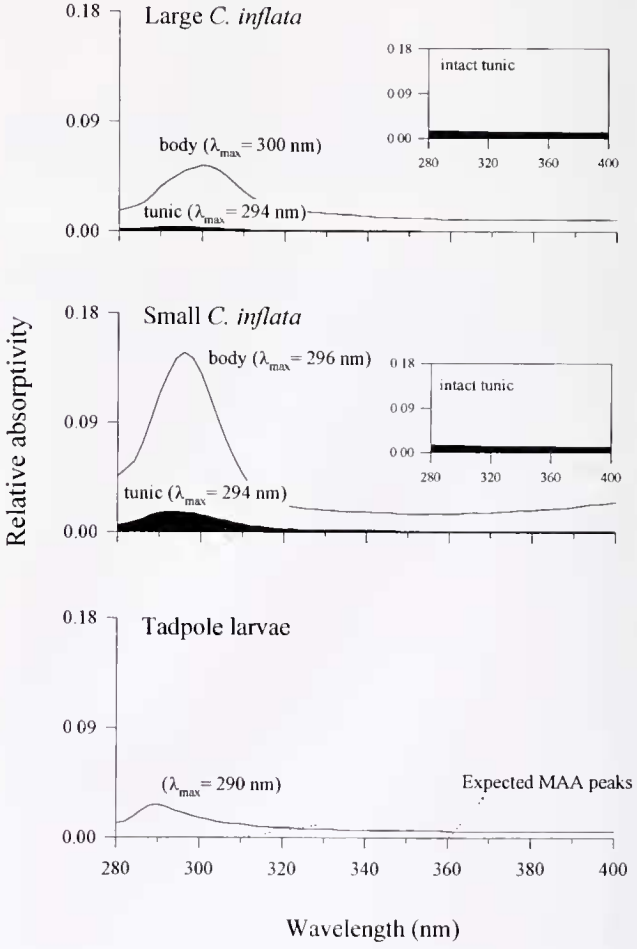

Figure 8. Relative absorptivities of *Corella inflata* extracts (80% methanol). Large individuals were >1.5 cm long; small individuals were <1.0 cm long. All gut contents were cleaned out during preparation to ensure that only *C. inflata* compounds were measured in the body samples. The area of normal mycosporine-like amino acid (MAA) absorbance peaks is indicated. Inset graphs are absorptivities for intact whole tunic. λ_{max} are maximum absorptivity peaks. See the text for absorptivity calculations.

higher, and it seems likely that those wavelengths also have a significant impact. The relative importance of UV-A, UV-B, and PAR to *C. inflata* populations merits further study. In our laboratory work, the UV/PAR ratios were higher than they would be under natural conditions. Many organisms possess DNA repair systems that are activated by short-wave visible light (reviewed by Mitchell and Karentz, 1993). If *C. inflata* has such systems, they may have been inoperable given the low PAR they received in our laboratory trials. The ideal way to test this possibility is to repeat the experiments under natural light in the field.

Whether UV effects are important for other ascidian species is unknown. Nearly 40 years ago, Endean (1961) suggested that pigmentation in the tunic of *Phallusia mamillata* protects that species from ultraviolet damage. To our knowledge, this has never been tested for *P. mamillata* or any other ascidian. Using a bacterial dosimeter, Karentz and Lutze (1990) detected significant UV-B radiation to 10 m and documented effects of irradiation to 20 and 30 m in Antarctic waters. Depth of UV penetration will depend on local conditions. However, in moderately clear, shallow waters, it is highly likely that UV will have significant ecological effects that may be detectable in life histories of the species that persist there.

Acknowledgments

We thank Steve Heywood and Gene McKeen for logistical assistance. Charley Lambert, Gretchen Lambert, Richard Cloney, and Jody Stecher provided unpublished information and/or valuable discussion and insight. We are also indebted to Deneb Karentz for patient guidance and suggestion. We appreciate the very helpful comments of two anonymous reviewers. The owners, staff, and residents of Anacortes, Cap Sante, Anchor Cove, and Skyline Marinas provided access to and use of their facilities. This work was supported by NSF grants OCE-9340328, USE-9650124, and USE-9151453.

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